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16TH PLANT DEVELOPMENT WORKSHOP

Saturday, November 9, 1985
McMaster University, Hamilton, Ontario

Preliminary Schedule

8:30 - 9:15	Registration, coffee
9:15 - 12:15	Contributed papers, special presentation on neutron activation analysis of elements found in biological tissues, tour of McMaster nuclear reactor
12:15 - 2:00	Buffet lunch, poster presentation, open house at the greenhouse, demonstration of cryogenic preparation of samples for SEM
2:00 - 4:30	Contributed papers
4:30 - 5:00	Reception

Presentations

Please submit an abstract typed in a box 17 cm. wide by 10 cm. deep. Give title, author, address and the text. Please indicate clearly if you wish this to be a poster or platform presentation.

For platform presentations indicate if 15 or 20 minutes is required for the talk plus questions.

Send Abstracts To

Irene Ockenden
Department of Biology
McMaster University
1280 Main Street West
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Telephone: (416) 525-9140 extension 4589 (J. Lott)

Information Requested

Please provide the following information as soon as possible:

- the number that will attend from your lab
- the number of posters and/or platform presentations you will give
- any requirements for accommodation

(16)

16TH PLANT DEVELOPMENT WORKSHOP NOVEMBER 9, 1985

MCMASTER UNIVERSITY

8:30 - 9:15 Registration, coffee
Psychology Building, Room 205

9:15 - 9:20 Announcements - Psychology, Room 155, John Lott

Session 1 - Contributed papers, chaired by Irene Ockenden

9:20 - 9:40 S.W. Armstrong and D. Francis
Differences in Cell Cycle Duration of Sister Cells in Secondary Root Meristems of *Cocos nucifera* L.

9:40 - 9:55 R.T. Riding and E. Mellerowicz
Cambial Reactivation in *Abies balsamea*

9:55 - 10:10 D. Davidson
Carrot Suspension Cultures: Loss of Ability to Proliferate

10:10 - 10:25 T. Finan
Mutants of *Rhizobium meliloti* Which Form Nodules Without Infection Threads.

10:25 - 10:40 Coffee, Psychology, Room.205

Session 2- Chaired by Ann Oaks
Psychology, Room 155

10:40 - 11:05 S. Landsberger
Application of Instrumental Neutron Activation Analyses to Plant and Soil Processes.

11:05 - 12:00 Conducted Tours of McMaster Nuclear Reactor and McMaster Greenhouse

12:10 - 13:00 Buffet Lunch:
Life Sciences Building, Room 103

13:00 - 14:00 1. Cryogenic Preparation of Samples for SEM
Demonstrated by Patrice Kerr.
Life Sciences Building, Room B126

2. Posters: Life Sciences Building, Room 102.

I. Ockenden,
Preservation of Seeds on Archaeological Sites.

A.R.Kermode and J.D.Bewley,
A Reprogramming of Protein Synthesis from a developmental to a Germinative Pattern is Induced by Premature Dessication of Castor Bean.

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V.Rastogi, L.A.Cass, and A.Oaks,
Control of Storage Protein Hydrolysis in Barley (Hordeum vulgare CV Perth) Endosperms.

A.Fyson and A.Oaks,
Corn Root Soil Sheaths; A Valuable Aid for the Study of Root / Micro-organism Interactions.

Session 3 - Contributed Papers:
Chaired by Douglas Davidson.
Psychology, Room 155

14:00 - 14:15 I. Ockenden and J.N.A. Lott
Neutron Activation Analysis in the Study of Minerals in Seeds.

14:15 - 14:35 R.A. Fletcher, G. Hofstra, N.K. Asare-Boamah, L. Krieg, and J. Gao
Triazoles - As Plant Multi-Protectants.

14:35 - 14:50 G. Sorger, D. Gooden, E.D. Earle and J. McKinnon
NADH Nitrate Reductase and NAD(P)H Nitrate Reductase in Genetic Variants and Regenerating Callus of Maize.

14:50 - 15:05 A.A. Sanchez-Burgos and N.G. Dengler
Facultative Anisophylly in Pentadenia Spp. (Gesneriaceae) I. Morphological Observations of Anisophylly in P. orientandina and Asophyly in P. crassicaulis.

15:05 - 15:20 J.P. Young and R.F. Horton
Heterophylly in Ranunculus flabellaris: The Effect of Abscisic Acid

15:20 - 15:40 C.M. Kampny and J.M. Canne
Floral Development in Agalinis fasciculata (Ell.) Raf. (Scrophulariaceae).

15:40 - 15:55 N.G. Dengler, P.A. Eastman and C.A. Peterson
Occurrence of a Suberin Lamella in Mestome Sheath Cell Walls of Grasses of Differing Photosynthetic Types.

15:55 - 16:10 C.A. Peterson
Does the Exodermal Casparian Band Constitute a Barrier to the Diffusion of Ions?

16:10 - 16:25 D.E. Enstone and C.A.Peterson
Comparison of the Movement of PTS and MBC in Bean Leaves

16:25 Closing Comments

16:30 - 17:00 Reception in Psychology 205

16TH PLANT DEVELOPMENT WORKSHOP

McMASTER UNIVERSITY

NOVEMBER 9, 1985

Differences in Cell Cycle Duration of Sister Cells in Secondary Root Meristems of Cocos nucifera L.

S.W. Armstrong and D. Francis, Dept. of Plant Science, University College, Cardiff, Wales, U.K.

Cell lengths of G1/G1 pairs of sister cells and in packets of mitotic cells were measured in secondary roots of C. nucifera. Due to symplastic growth cell length ratios of sister cells, over a cell cycle, remained constant for dwarf, hybrid and tall varieties of coconut; the mean cell length ratio, larger to smaller cell, was between 1.16:1 and 1.20:1. From the cell length ratios, mean differences in cell cycle duration for sister cells were calculated to lie between 0.22 and 0.27 of the average cell cycle duration.

Cumulative distributions of the cell length ratios of sister cells, when converted to differences in cell cycle duration and plotted on semi-log graph paper deviated from an exponential distribution. In 86 per cent of the sister cells studied, the larger cell of a sister pair divided first, however, in the remaining 14 per cent, half had the smaller cell of a sister pair dividing first while in the other half of the sister pairs, the cells were of equal length but one cell had entered mitosis ahead of its sister. These results support neither cell cycle models based on a cell sizer control per se nor on a random transition from inactive to active parts of the cell cycle.

CAMBIAL REACTIVATION IN ABIES BALSAMEA. R.T. Riding, E. Mellerowicz, Department of Biology, University of New Brunswick, Fredericton, N.B. E3B 6E1

Field grown trees are being sampled to investigate the transition between active growth, rest, quiescense, and reactivation of the vascular cambium. To date, 10 trees have been sampled bi-weekly throughout the reactivation period from March to June. Reactivation of the cambium was evident in the outer portion of the crown before the main bole. A decrease in staining intensity for total protein was evident before the onset of cell division at all positions within the tree; this was evident on 22 April. A conspicuous zone of xylem mother cells was evident on 7 May. Within the crown, xylem maturation proceeded from tips to bases of branches. Throughout the study period, proteins stained less intensely at the base of the tree and branches than at the tips. Changes in nuclear size and composition are also being investigated.

D. Davidson, Biology Department, McMaster University, Hamilton, Ont. Carrot Suspension Cultures: Loss of Ability to Proliferate.

Carrot (Daucus carota) cell suspension cultures were established from petiole explants; they were maintained on liquid M and S medium containing 0.1 mg/l 2,4-D and 0.2 mg/l kinetin. Cell suspensions were routinely sieved through 280 µm screens to remove the largest cell clusters. These large clusters, or aggregates, of cells soon reappear in cultures, however. The origin of the large clusters was studied by: a) determining the frequency of mitoses in cells of different sizes; b) analysing the morphogenesis of the clusters of cells. Two cell types were present: 1) small cells, 8-20 µm in diameter; 2) large cells were 30-180 µm in length and 15-20 µm in diameter or were barrel-shaped cells, 80-120 µm in diameter. 94.1% of all mitoses were in small cells: their mitotic index was 9.52. In large cells, mitotic index was 0.3. These values are based on a sample of 3000 cells. Furthermore, the 6 large cells in division were 23-39 µm in diameter; larger cells were never seen in division. It appears that large cells are not a self-generating subpopulation: instead, they are descended from small cells that have stopped dividing but have continued to expand. Analysis of clusters of 8-16 cells indicates that all cells were about the same size. It appears that a cell may divide 3-4 times, giving 8-16 cells, and then all cells stop dividing and expand. These results will be related to the evidence that two physiologically distinct populations are present in carrot suspension cultures.

Mutants of Rhizobium meliloti which form nodules without infection threads. Turlough Finan, Dept. of Biology, McMaster University, Hamilton, Onrario L8S 4K1.
Mutations at a Fix locus in R.meliloti result in a conditional insensitivity to a monoclonal antibody to the bacterial cell surface, and resistance to several phages apparently because of a defect in the synthesis of a wild type exopolysaccharide (Exo^-).On alfalfa the mutants do not induce marked root hair curling, but penetrate the epidermis directly, forming nodules that contain no visible infection threads or "bacteriods". The nodules, which fail to fix N2, have a few bacteria in the intercellular spaces only and not within the nodule cells. Thus, infection thread formation is not essential for cell proliferation and nodule formation, which are here evidently induced by a bacterial signal at a distance. The Fix locus is located on a megaplasmid in R.meliloti which is distinct from the known nod nif megaplasmid.

Preservation of Seeds on Archaeological Sites.

Irene Ockenden. Biology Department, McMaster University.

Prehistoric and early historic Indians of Ontario cultivated corn, beans, squash, sunflower and tobacco. Traces of these cultigens are found in middens and houses on archaeological sites. Corn kernels and beans, mainly as fragments, are commonly found while squash, sunflower and tobacco finds are rare. The preserved fragments appear black and brittle and are considered to be charred or carbonized. Investigation of the internal structure of preserved corn and beans from the Thorold Site by scanning electron microscopy showed considerable variation in the appearance of the specimens. None of the fragments showed a similarity to freshly charred modern corn kernels and bean seeds. The structure of the corn kernels suggested decay had preceeded charring. Beans from the site appear to have been preserved at least partly by mineralization rather than only charring.

A REPROGRAMMING OF PROTEIN SYNTHESIS FROM A DEVELOPMENTAL TO A GERMINATIVE PATTERN IS INDUCED BY PREMATURE DESICCATION OF CASTOR BEAN.

Allison R. Kermode and J. Derek Bewley. Department of Botany, University of Guelph, Guelph, Ontario, N1G 2W1, Canada.

Seeds removed from the capsule at 50 days after pollination (DAP) and later will germinate when placed in water, whereas developing seeds at 25-45 DAP require a desiccation period before germination can occur. This switch from development to germination elicited by premature drying is mirrored by a redirection in metabolism within the endosperm of the castor bean seed. Isolated endosperms of 30 and 40 DAP seeds, which have not completed all developmental events, are tolerant of premature drying and will respond upon subsequent rehydration in a manner which is characteristic of an endosperm from a germinating mature seed. In particular, the pattern of soluble and insoluble protein synthesis upon rehydration of dried 30 and 40 DAP seeds is identical to that of hydrated mature endosperms: proteins characteristic of development cease to be synthesized while those associated with germination and growth are then produced. The induction of enzymes essential to the post-germinative (growth) phase of seedling development occurs as a consequence of premature drying. Isocitrate lyase is virtually absent from developing endosperms, but those dried at 40 DAP are induced to synthesize the enzyme at levels comparable to normal germinated seeds. LeuNAase, a proteolytic enzyme, responds to drying in a similar manner.

CONTROL OF STORAGE PROTEIN HYDROLYSIS IN BARLEY (*HORDEUM VULGARE* CV PERTH) ENDOSPERMS

Vipin Rastogi, Leslie A. Cass, and Ann Oaks. Department of Biology, McMaster University, Hamilton, Ont., Canada L8S 4K1.

Prolamins--the major storage proteins in barley endosperms--are hydrolyzed to small peptides and amino acids during germination. The complete hydrolysis of endosperm reserves is dependent on enzymes synthesized in response to gibberellic acid (GA_3). For example, GA_3 leads to the induction of α-amylase and a carboxypeptidase. The appearance of these two enzymes is inhibited by cordycepin and cycloheximide. We suspect that the carboxypeptidase is a scavenger protease not related specifically to the initial hydrolysis of hordein. To determine whether there was a specific protease, we examined the soluble products released from the initial hydrolysis of prolamins. Proteins were prepared from dry endosperms and endosperms incubated for 24 h in a buffered media in the presence or absence of GA_3, treated with SDS-PAGE electrophoresis, and then transblotted to nitrocellulose. The hordein-derived protein bands were identified by IgG prepared against urea denatured hordeins. Several bands (3 major and 6-7 minor) were detected on Western blots of the dry seed sample. Two other bands ($\sim$25 and $\sim$35 kD) were noted in the minus GA_3 samples. The appearance of 2 minor bands ($\sim$38 and $\sim$40 kD) and increased intensity of several other bands in the 10-15 kD size range was evident in the plus GA_3 samples. The addition of cycloheximide or cordycepin prevented the appearance of the 25, 35, 38 and 40 kD bands as well as the lower molecular weight bands associated with the GA_3 treatment. The results are consistent with the idea that the synthesis of a specific protease which is independent of GA_3 is involved in the early steps of hydrolysis. However a *de novo* synthesis of this enzyme appears to be required for the appearance of this activity. Other protease(s) which required both *de novo* synthesis and GA_3 are necessary for the overall hydrolysis of hordein.

CORN ROOT SOIL SHEATHS - A VALUABLE AID TO THE STUDY OF ROOT-MICROORGANISM INTERACTIONS. Andrew Fyson & Ann Oaks, Department of Biology, McMaster University, Hamilton, Ontario.

The development of soil sheaths was observed in primary roots of corn. Zea mays (cv. W64A x W183E) was sown in a controlled environment cabinet (28°C; 16h/8h, light/dark cycle) in field soils which had been under continuous corn or continuous alfalfa for five years. At 50h from sowing, seedlings were carefully excavated and tapped to remove loose soil. The remaining soil was washed off with vigorous shaking and the sheath weight estimated by difference. Roots in alfalfa soil retained 1.9 mg soil/mm root whereas corn soil roots retained only 0.5 mg soil/mm root. Gamma irradiation had no significant effect on the size or resilience of soil sheaths. Therefore, in these soils, microorganisms do not play an essential role in root sheath initiation. Fine soil fractions (particle size <425 μm) produced more resilient sheaths than courser fractions (particle size >425 μm). The physical characteristics of the soil therefore determine the resilience of the sheath. The stability and reproducibility of these structures makes them a valuable tool for studies of root colonisation by microorganisms and of effects of microorganisms on early plant development.

Neutron Activation Analysis in the Study of Minerals in Seeds.
I. Ockenden and J.N.A. Lott. Biology Department, McMaster University

Neutron activation analysis of unashed embryo tissues was carried out to determine whether absolute or relative differences in the storage mineral concentrations were responsible for observed differences in the ash characteristics of various embryos. The ashes differed in colour, consistency and pH and as well in the ease with which calcium could be extracted from the ash. It was found that calcium was bound in a relatively insoluble form in the ashes of all tested members of the Family Cucurbitaceae and also in preparations of castor bean endosperm plus embryo tissues. The binding was the result of a lower potassium to phosphorus ratio in these embryos as compared to embryos which did not show calcium binding. Solid state interactions of the salts produced during ashing apparently result in the formation of various compounds depending on the relative amount of the minerals initially present in the embryos.

TRIAZOLES - AS PLANT MULTI-PROTECTANTS. R.A. Fletcher, G. Hofstra, N.K. Asare-Boamah, Lori Krieg and J. Gao. Department of Environmental Biology, University of Guelph, Guelph, Ontario N1G 2W1.

Recently, several triazole derivatives have been developed for use as either fungicides or plant growth regulators. However in varying degrees they exhibit both fungicidal and plant growth regulating properties. In addition to these properties, the triazoles, triadimefon and S-3307 also protect plants from injury due to drought, heat, chilling and ozone. Preliminary evidence indicates that the multiple effects of the triazole derivates are mediated by regulating the isoprenoid pathway and thereby shifting the balance of important plant hormones in the pathway. Besides its agricultural implications, the triazole derivatives provides the biologist with a powerful tool for investigating how a plant adjusts its hormonal balance in response to environmental stress.

lNADH nitrate reductase and NAD(P)H nitrate reductase in genetic variants and regenerating callus of maize

George Sorger 1 , Dinsdale O . Gooden 1,Elizabeth D. Earle 2 and Joanne McKinnon 1

1 Department of Biology, McMaster University, Hamilton, Ontario,L8S4K1, Canada
2 Department of Plant Breeding and Biometry, New York State College of Agriculture and Life Sciences, Cornell University, Ithaca, N. Y. 14853 ,USA

Abstract

A genetic variant of maize has been isolated from the second generation progeny of a selfed Mu (Robertson's mutator) containing strain. the variant has a relatively low level of NAD(P)H nitrate reductase (E.C. 1.6.6.2.) in scutellar and root extracts, the leaf NADH nitrate reductase is normal. Extracts of undifferentiated and regenerating callus in two different strains of corn contain both NADH and NAD(P)H nitrate reductase activities, as do extracts of scutellum and root.Leaf extracts contain NADH nitrate reductase activity and little or no NAD(P)H nitrate reductase activity. The relative content of NADH and NAD(P)H nitrate reductase activity in the scutellum is influenced significantly gy the age of the seedling.

FACULTATIVE ANISOPHYLLY IN PENTADENIA SPP. (GESNERIACEAE). I. MORPHOLOGICAL OBSERVATIONS OF ANISOPHYLLY IN P. ORIENTANDINA AND ASOPHYLY IN P. CRASSICAULIS. Arturo A. Sanchez-Burgos and Nancy G. Dengler, Dept. of Botany, University of Toronto.

Both anisophyllous and isophyllous species may be found within the same genus in the Gesneriaceae. Morphological studies of leaf development in representative species of the genus Pentadenia show that, despite variability in the degree of anisophylly expressed at maturity, leaf development is characterized by: 1) a period during and shortly after leaf inception when leaves of a pair are slightly unequal in size, and 2) a period of leaf expansion during which either both leaves expand fairly equally (resulting in an isophyllous node) or the rate of growth of one leaf of the pair is reduced, followed by precocious maturation of that leaf (resulting in an anisophyllous node). We conclude that, in agreement with previous work on habitually anisophyllous species, leaf size usually differs from inception in facultative anisophyllous species. Plasticity of leaf size is expressed through divergent or similar growth at later developmental stages.

HETEROPHYLLY IN RANUNCULUS FLABELLARIS: THE EFFECT OF ABSCISIC ACID. Jane P. Young and Roger F. Horton. Dept. of Botany, University of Toronto, Toronto, Ontario, Toronto, Ontario, M5S 1A1 and Dept. of Botany, University of Guelph, Guelph, Ontario, N1G 2W1

R. flabellaris, the yellow water-crowfoot, is a semi-aquatic angiosperm. It exhibits a striking heterophylly between terrestrial and submerged leaves. Emergent shoots have entire, tri-lobed leaves whereas leaves produced underwater are highly dissected. Variation in leaf form in plants grown under controlled conditions was assessed using image analysis techniques, and the degree of heterophylly was expressed as the Heterophylly Index (HI). Leaves produced after submergence of plants previously grown terrestrially demonstrated progressively greater HI values, ultimately resulting in filiform leaves. Anatomical and ultrastructural studies also revealed differences between submerged and terrestrial leaf types.

When plants were submerged in 25 uM abscisic acid (ABA) solution, the transition from the terrestrial to the submerged leaf form was completely suppressed and instead, terrestrial-form leaves were produced. In addition to leaf form, ABA treated leaves have anatomical structures similar to those of terrestrial leaves. Detailed structural analyses are presently being done to quantify these results.

This study provides evidence that the environmental factors that influence the expression of heterophylly in this species, including internal structure, may act through endogenous plant growth regulators.

Floral development in Agalinis fasciculata (Ell.) Raf. (Scrophulariaceae)

C.M. Kampny and J.M. Canne, Dept. of Botany, Univ. of Guelph

The following project is part of an investigation to determine the developmental origins of the various floral forms existing among North and South American taxa of Agalinis and allies, and to observe the sequence of appearance of specific and generic characters. The floral development of Agalinis fasciculata was observed with the dissecting microscope and with the SEM. Calyx lobe initiation occurred first on the posterior side of the floral apex. In the corolla, the three anterior lobes arose first, almost simultaneously with the four stamen primordia. The two posterior corolla lobes did not appear until after the gynoecium had been initiated as two raised areas forming a zygomorphic ring. The early growth of both the calyx and the corolla tube was asymmetric. The stamens maintained their pattern of larger anterior filaments and thecae throughout their development. Intercalary growth closed the gynoecium and then resulted in an elongate style with a two- or fourlobed stigma which was later covered with stigmatic trichomes. Ridges growing upwards and inwards inside the ovary formed a septum with a placenta on each side bearing many small ovules.

time: 20 minutes

OCCURENCE OF A SUBERIN LAMELLA IN MESTOME SHEATH CELL WALLS OF GRASSES OF DIFFERING PHOTOSYNTHETIC TYPES. Nancy G. Dengler, Dept. of Botany, University of Toronto, P. Ann Eastman and Carol A. Peterson, Dept. of Biology, University of Waterloo.

A mestome sheath of elongated, thick-walled cells surrounds most of the longitudinal veins in grass leaves. In conjunction with studies using PTS as an apoplastic tracer, we have undertaken an ultrastructural survey of 8 grass species, belonging to 4 photosynthetic types and 5 major taxonomic groups. Our observations show that a suberin lamella is present in at least the outer tangential and radial walls of mestome sheath cells surrounding the major veins in each species. In contrast, in minor veins, a continuous mestome sheath having a suberin lamella is present only in C3 and NADP-ME species. Transverse veins of all species examined, except NADP-ME species, also lacked a recognizable mestome sheath and any indication of a suberin lamella.

These results indicate that mestome sheath cells potentially provide an apoplastic barrier to water movement from major veins to mesophyll only. The observed leakage of PTS tracer from major veins may occur between adjacent suberin lamallae in radial walls or reflect the large proportion of fluid moving through major as compared with minor veins. Since the mestome sheath cells of NADP-ME grasses also function as Kranz cells, the role of the suberin lamella of these cell walls may be to reduce apoplastic leakage of HCO_3^-.

DOES THE EXODERMAL CASPARIAN BAND CONSTITUTE A BARRIER TO THE DIFFUSION OF IONS? Carol A. Peterson, Department of Biology, University of Waterloo.

Results of histochemical tests and the diffusion pattern of apoplastic dyes have indicated the presence of an impermeable Casparian band in hypodermal layers of roots of many species. While on sabbatical leave, in the laboratory of M. Pitman at the University of Sydney, I tested the permeability of the onion root exodermal Casparian band to sulfate ions to determine whether the previously-observed dye blockage reflects the movement of ions. The hypothesis was that if the exodermal Casparian band were impermeable to sulfate, the sulfate free space should equal the free space of the epidermis but if the Casparian band were permeable to sulfate, the sulfate free space should equal the free space of the epidermis plus cortex. The sulfate free spaces were measured by an elution method in (a) bisected root segments with their steles removed, and (b) root segments with cut ends sealed with sticky wax (10 segments, 20 mm long). The free space in the former was 10.13 µL while the free space in the latter was 2.80 µL. Total wall volumes were 16.07 µL for the epidermis plus cortex, and 4.21 µL for the epidermis alone. Thus, the sulfate free space was about 65% of the total wall volume in each case. The ratio of sulfate free spaces (a/b above) = 3.62. The ratio of wall volumes epidermis + cortex/epidermis = 3.81. The results are therefore consistent with the hypothesis that the exodermal Casparian band of onion is impermeable to the diffusion of sulfate ions.

COMPARISON OF THE MOVEMENT OF PTS AND MBC IN BEAN LEAVES. Daryl E. Enstone and Carol A. Peterson, Department of Biology, University of Waterloo.

It was previously noted that the transport of the fluorescent dye PTS, differed from the transport of the fungicide, MBC, in plant roots. This observation ultimately led to the realization that PTS is euapoplastic (i.e. confined to plant cell walls and unable to penetrate membranes) whereas MBC is pseudoapoplastic (i.e. able to diffuse across membranes). When applied to roots, PTS was filtered out by the Casparian bands but the MBC diffused into the stele and was carried upward in the transpiration stream.

The transport patterns of these two chemicals in leaves are also strikingly different. PTS produces a fluorescent spotted appearance in the primary leaves. These spots are formed as the dye is washed out of the minor veins into the mesophyll in the vicinity of the vein endings. Further flow of water out of the veins localizes PTS into spots and prevents it from diffusing evenly through the tissue. In contrast, MBC is initially distributed evenly through the leaf blade (as determined by autoradiography and fungal bioassay); however with time, it accumulates in phytotoxic concentrations in the margins and tips of the leaves. This accumulation is not affected by diurnal stomatal cycling nor by inhibition of phloem transport by nickel. Currently we are in the process of locating a fluorescent dye which will mimic the transport pattern of MBC so that we can better visualize its movement.